

SEQUENCE LISTING

<110> Lee, Arthur M.E.
Jain, Mukesh
Watanabe, Masafumi

<120> IN VITRO DIFFERENTIATION OF VASCULAR SMOOTH MUSCLE
CELLS AND REAGENTS RELATED THERETO

<130> APV-382.01

<140> 09/181,311

<141> 1998-10-28

<150> 60/063,363

<151> 1997-10-28

<150> 60/080,420

<151> 1998-04-02

<150> 60/096,685

<151> 1998-08-14

<160> 23

<170> PatentIn Ver. 2.0

<210> 1

<211> 153

<212> DNA

<213> Murine

<220>

<223> Clone WO11

<400> 1

agtgggtgat gtggggaagc cttctgtgaa aacgtggaag gaccccttct gtgtgtgtgt 60
gccgatgaga accaggagta cagcccatg actgggcagt gtcgctctcg ggtcacggaa 120
gattcagggtg tggatcgta gccatggcga aga 153

<210> 2

<211> 143

<212> DNA

<213> Rattus

<220>

<223> Clone LTBP-1

<400> 2

agtgggtgat gtggggaggc tttctgtgaa aatgtggaag ggtcccttct gtgcgtgtgt 60
gccgatgaga accaggagta cagcccatg actgggcagt gtcgctcccg ggctactgaa 120
gattcagggtg tggatcgta gcc 143

<210> 3

<211> 1128

<212> PRT

<213> Homo sapiens



<220>

<223> ACLP

<400> 3

Met Ala Ala Val Arg Gly Ala Pro Leu Leu Ser Cys Leu Leu Ala Leu
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Leu Ala Leu Cys Pro Gly Gly Arg Pro Gln Thr Val Leu Thr Asp Asp
 20 25 30

Glu Ile Glu Glu Phe Leu Glu Gly Phe Leu Ser Glu Leu Glu Pro Glu
 35 40 45

Pro Arg Glu Asp Asp Val Glu Ala Pro Pro Pro Pro Glu Pro Thr Pro
 50 55 60

Arg Val Arg Lys Ala Gln Ala Gly Gly Lys Pro Gly Pro Gly Thr Ala
 65 70 75 80

Ala Glu Val Pro Pro Glu Lys Thr Lys Asp Lys Gly Lys Lys Gly Lys
 85 90 95

Lys Asp Lys Gly Pro Lys Val Pro Lys Glu Ser Leu Glu Gly Ser Pro
 100 105 110

Arg Pro Pro Lys Lys Gly Lys Glu Lys Pro Pro Lys Ala Thr Lys Lys
 115 120 125

Pro Lys Glu Lys Pro Pro Lys Ala Thr Lys Lys Pro Lys Glu Glu Pro
 130 135 140

Pro Lys Ala Thr Lys Lys Pro Lys Glu Lys Lys Ala Thr Lys Lys Pro
 145 150 155 160

Pro Ser Gly Lys Arg Pro Pro Ile Leu Ala Pro Ser Glu Thr Leu Glu
 165 170 175

Trp Pro Leu Pro Pro Pro Pro Ser Pro Gly Pro Glu Glu Leu Pro Gln
 180 185 190

Glu Gly Gly Ala Pro Leu Ser Asn Asn Trp Gln Asn Pro Gly Glu Glu
 195 200 205

Thr His Val Glu Ala Gln Glu His Gln Pro Glu Pro Glu Glu Glu Thr
 210 215 220

Glu Gln Pro Thr Leu Asp Tyr Asn Ile Glu Arg Glu Asp Tyr Glu Asp
 225 230 235 240

Phe Glu Tyr Ile Arg Arg Gln Lys Gln Pro Arg Pro Pro Pro Ser Arg
 245 250 255

Arg Arg Arg Pro Glu Arg Val Trp Pro Glu Pro Pro Glu Glu Lys Ala
 260 265 270

Pro Ala Pro Ala Pro Glu Glu Arg Ile Glu Pro Pro Val Lys Pro Leu
 275 280 285

Leu Pro Pro Leu Pro Pro Asp Tyr Gly Asp Gly Tyr Val Ile Pro Asn
 290 295 300
 Tyr Asp Asp Met Asp Tyr Gly Pro Pro Pro Pro Gln Lys Pro Asp Ala
 305 310 315 320
 Glu Arg Gln Thr Asp Glu Glu Lys Glu Glu Leu Lys Lys Pro Lys Lys
 325 330 335
 Glu Asp Ser Ser Pro Lys Glu Glu Thr Asp Lys Trp Ala Val Glu Lys
 340 345 350
 Gly Lys Asp His Lys Glu Pro Arg Lys Gly Glu Glu Leu Glu Glu Glu
 355 360 365
 Trp Thr Pro Thr Glu Lys Val Lys Cys Pro Pro Ile Gly Met Glu Ser
 370 375 380
 His Arg Ile Asn Gln Ile Arg Ala Ser Ser Met Leu Arg His Gly Leu
 385 390 395 400
 Gly Ala Gln Arg Gly Arg Leu Asn Met Gln Thr Gly Ala Thr Glu Asp
 405 410 415
 Asp Tyr Tyr Asp Gly Ala Trp Cys Ala Glu Asp Asp Ala Arg Thr Gln
 420 425 430
 Trp Ile Glu Val Asp Thr Arg Arg Thr Thr Arg Phe Thr Gly Val Ile
 435 440 445
 Thr Gln Gly Arg Asp Ser Ser Ile His Asp Asp Phe Val Thr Thr Phe
 450 455 460
 Phe Phe Ser Asn Asp Ser Gln Thr Trp Val Met Tyr Thr Asn Gly Tyr
 465 470 475 480
 Glu Glu Met Thr Phe His Gly Asn Val Asp Lys Asp Thr Pro Val Leu
 485 490 495
 Ser Glu Leu Pro Glu Pro Val Val Ala Arg Phe Ile Arg Ile Tyr Pro
 500 505 510
 Leu Thr Trp Asn Gly Ser Leu Cys Met Arg Leu Glu Val Leu Gly Cys
 515 520 525
 Ser Val Ala Pro Val Tyr Ser Tyr Tyr Ala Gln Asn Glu Val Val Asp
 530 535 540
 Asp Leu Asp Phe Arg His His Ser Tyr Lys Asp Met Arg Gln Leu Met
 545 550 555 560
 Lys Val Val Asn Glu Cys Pro Thr Ile Thr Arg Thr Tyr Ser Leu
 565 570 575
 Gly Lys Ser Ser Arg Gly Leu Lys Ile Tyr Ala Met Glu Ile Ser Asp
 580 585 590

Asn Pro Gly Glu His Glu Leu Gly Glu Pro Glu Phe Arg Tyr Thr Ala
 595 600 605
 Gly Ile His Gly Asn Glu Val Leu Gly Arg Glu Leu Ile Leu Met Gln
 610 615 620
 Tyr Leu Cys Arg Glu Tyr Arg Asp Gly Asn Pro Arg Val Arg Ser Leu
 625 630 635 640
 Val Gln Asp Thr Arg Ile His Leu Val Pro Ser Leu Asn Pro Asp Gly
 645 650 655
 Tyr Glu Val Ala Ala Gln Met Gly Ser Glu Phe Gly Asn Trp Ala Leu
 660 665 670
 Gly Leu Trp Thr Glu Glu Gly Phe Asp Ile Phe Glu Asp Phe Pro Asp
 675 680 685
 Leu Asn Ser Val Leu Trp Gly Ala Glu Glu Phe Val Pro Tyr Arg Val
 690 695 700
 Pro Asn Asn Asn Leu Pro Ile Pro Glu Arg Tyr Leu Ser Pro Asp Ala
 705 710 715 720
 Thr Val Ser Thr Glu Val Arg Ala Ile Ile Ala Trp Met Glu Lys Asn
 725 730 735
 Pro Phe Val Leu Gly Ala Asn Leu Asn Gly Gly Glu Arg Leu Val Ser
 740 745 750
 Tyr Pro Tyr Asp Met Ala Arg Thr Pro Thr Gln Glu Gln Leu Leu Ala
 755 760 765
 Ala Ala Met Ala Ala Ala Glu Gly Glu Asp Glu Val Ser Glu Ala Gln
 770 775 780
 Glu Thr Pro Asp His Ala Ile Phe Arg Trp Leu Ala Ile Ser Phe Ala
 785 790 795 800
 Ser Ala His Leu Thr Leu Thr Glu Pro Tyr Arg Gly Gly Cys Gln Ala
 805 810 815
 Gln Asp Tyr Thr Gly Gly Met Gly Ile Val Asn Gly Ala Lys Trp Asn
 820 825 830
 Pro Arg Thr Gly Thr Ile Asn Asp Phe Ser Tyr Leu His Thr Asn Cys
 835 840 845
 Leu Glu Leu Ser Phe Tyr Ile Asp Lys Phe Pro His Glu Ser Glu Leu
 850 855 860
 Pro Arg Glu Trp Glu Asn Asn Lys Glu Ala Leu Leu Thr Phe Met Glu
 865 870 875 880
 Gln Val His Arg Gly Ile Lys Gly Val Val Thr Asp Glu Gln Gly Ile
 885 890 895

Pro Ile Ala Asn Ala Thr Ile Ser Val Ser Gly Ile Asn His Gly Val
900 905 910

Lys Thr Ala Ser Gly Gly Asp Tyr Trp Arg Ile Leu Asn Pro Gly Glu
915 920 925

Tyr Arg Val Thr Ala Glu Gly Tyr Thr Pro Ser Ala Lys Thr Cys Asn
930 935 940

Val Asp Tyr Asp Ile Gly Ala Thr Gln Cys Asn Phe Ile Leu Ala Arg
945 950 955 960

Ser Asn Trp Lys Arg Ile Arg Glu Ile Met Ala Met Asn Gly Asn Arg
965 970 975

Pro Ile Pro His Ile Asp Pro Ser Arg Pro Met Thr Pro Gln Gln Arg
980 985 990

Arg Leu Gln Gln Arg Arg Leu Gln His Arg Leu Arg Leu Arg Ala Gln
995 1000 1005

Met Arg Leu Asn Ala Thr Thr Thr Leu Gly Pro His Thr Val Pro Pro
1010 1015 1020

Thr Leu Pro Pro Ala Pro Ala Thr Thr Leu Ser Thr Thr Ile Glu Pro
1025 1030 1035 1040

Trp Gly Leu Ile Pro Pro Thr Thr Ala Gly Trp Glu Glu Ser Glu Thr
1045 1050 1055

Glu Thr Tyr Thr Glu Val Val Thr Glu Phe Gly Thr Glu Val Glu Pro
1060 1065 1070

Glu Phe Gly Thr Lys Val Glu Pro Glu Phe Glu Thr Gln Leu Phe Glu
1075 1080 1085

Thr Gln Leu Glu Pro Glu Phe Glu Glu Glu Glu Glu Glu Lys Glu
1090 1095 1100

Glu Glu Ile Ala Thr Gly Gln Ala Phe Pro Phe Thr Thr Val Glu Thr
1105 1110 1115 1120

Tyr Thr Val Asn Phe Gly Asp Phe
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<212> PRT
<213> Murine

<220>
<223> ACLP

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 Glu Ile Glu Glu Phe Leu Glu Gly Phe Leu Ser Glu Leu Glu Thr Gln
 35 40 45
 Ser Pro Pro Arg Glu Asp Asp Val Glu Val Gln Pro Leu Pro Glu Pro
 50 55 60
 Thr Gln Arg Pro Arg Lys Ser Lys Ala Gly Gly Lys Gln Arg Ala Asp
 65 70 75 80
 Val Glu Val Pro Pro Glu Lys Asn Lys Asp Lys Glu Lys Lys Gly Lys
 85 90 95
 Lys Asp Lys Gly Pro Lys Ala Thr Lys Pro Leu Glu Gly Ser Thr Arg
 100 105 110
 Pro Thr Lys Lys Pro Lys Glu Lys Pro Pro Lys Ala Thr Lys Lys Pro
 115 120 125
 Lys Glu Lys Pro Pro Lys Ala Thr Lys Lys Pro Lys Glu Lys Pro Pro
 130 135 140
 Lys Ala Thr Lys Lys Pro Lys Glu Lys Lys Ala Thr Lys Arg Pro Ser
 145 150 155 160
 Ala Gly Lys Lys Phe Ser Thr Val Ala Pro Leu Glu Thr Leu Asp Arg
 165 170 175
 Leu Leu Pro Ser Pro Ser Asn Pro Ser Ala Gln Glu Leu Pro Gln Lys
 180 185 190
 Arg Asp Thr Pro Phe Pro Asn Ala Trp Gln Gly Gln Gly Glu Glu Thr
 195 200 205
 Gln Val Glu Ala Lys Gln Pro Arg Pro Glu Pro Glu Glu Glu Thr Glu
 210 215 220
 Met Pro Thr Leu Asp Tyr Asn Ile Glu Lys Glu Asp Tyr Glu Asp Phe
 225 230 235 240
 Glu Tyr Ile Arg Arg Gln Lys Gln Pro Arg Pro Thr Pro Ser Arg Arg
 245 250 255
 Arg Leu Trp Pro Glu Arg Pro Glu Glu Lys Thr Glu Glu Pro Glu Glu
 260 265 270
 Arg Lys Glu Val Glu Pro Pro Leu Lys Pro Leu Leu Pro Pro Asp Tyr
 275 280 285
 Gly Asp Ser Tyr Val Ile Pro Asn Tyr Asp Asp Leu Asp Tyr Pro His
 290 295 300
 Pro Pro Pro Gln Lys Pro Asp Val Gly Gln Glu Val Asp Glu Glu Lys
 305 310 315 320

Glu Glu Met Lys Lys Pro Lys Lys Glu Gly Ser Ser Pro Lys Glu Asp
 325 330 335
 Thr Glu Asp Lys Trp Thr Val Glu Lys Asn Lys Asp His Lys Gly Pro
 340 345 350
 Arg Lys Gly Glu Glu Leu Glu Glu Trp Ala Pro Val Glu Lys Ile
 355 360 365
 Lys Cys Pro Pro Ile Gly Met Glu Ser His Arg Ile Asn Gln Ile Arg
 370 375 380
 Ala Ser Ser Met Leu Arg His Gly Leu Gly Ala Gln Arg Gly Arg Leu
 385 390 395 400
 Asn Met Gln Ala Gly Ala Asn Glu Asp Asp Tyr Tyr Asp Gly Ala Trp
 405 410 415
 Cys Ala Glu Asp Glu Ser Gln Thr Gln Trp Ile Glu Val Asp Thr Arg
 420 425 430
 Arg Thr Thr Arg Phe Thr Gly Val Ile Thr Gln Gly Arg Asp Ser Ser
 435 440 445
 Ile His Asp Asp Phe Val Thr Thr Phe Phe Phe Ser Asn Asp Ser Gln
 450 455 460
 Thr Trp Val Met Tyr Thr Asn Gly Tyr Glu Glu Met Thr Phe Tyr Gly
 465 470 475 480
 Asn Val Asp Lys Asp Thr Pro Val Leu Ser Glu Leu Pro Glu Pro Val
 485 490 495
 Val Ala Arg Phe Ile Arg Ile Tyr Pro Leu Thr Trp Asn Gly Ser Leu
 500 505 510
 Cys Met Arg Leu Glu Val Leu Gly Cys Pro Val Thr Pro Val Tyr Ser
 515 520 525
 Tyr Tyr Ala Gln Asn Glu Val Val Asp Ser Leu Asp Phe Arg His His
 530 535 540
 Ser Tyr Lys Asp Met Arg Gln Leu Met Lys Ala Val Asn Glu Glu Cys
 545 550 555 560
 Pro Thr Ile Thr Arg Thr Tyr Ser Leu Gly Lys Ser Ser Arg Gly Leu
 565 570 575
 Lys Ile Tyr Ala Met Glu Ile Ser Asp Asn Pro Gly Asp His Glu Leu
 580 585 590
 Gly Glu Pro Glu Phe Arg Tyr Thr Ala Gly Ile His Gly Asn Glu Val
 595 600 605
 Leu Gly Arg Glu Leu Ile Leu Met Gln Tyr Leu Cys Gln Glu Tyr Arg
 610 615 620

Asp Gly Asn Pro Arg Val Arg Asn Leu Val Gln Asp Thr Arg Ile His
 625 630 635 640
 Leu Val Pro Ser Leu Asn Pro Asp Gly Tyr Glu Val Ala Ala Gln Met
 645 650 655
 Gly Ser Glu Phe Gly Asn Trp Ala Leu Gly Leu Trp Thr Glu Glu Gly
 660 665 670
 Phe Asp Ile Phe Glu Asp Phe Pro Asp Leu Asn Ser Val Leu Trp Ala
 675 680 685
 Ala Glu Glu Phe Val Pro Tyr Arg Val Pro Asn Asn Asn Leu Pro Ile
 690 695 700
 Pro Glu Arg Tyr Leu Ser Pro Asp Ala Thr Val Ser Thr Glu Val Arg
 705 710 715 720
 Ala Ile Ile Ser Trp Met Glu Lys Asn Pro Phe Val Leu Gly Ala Asn
 725 730 735
 Leu Asn Gly Gly Glu Arg Leu Val Ser Tyr Pro Tyr Asp Met Ala Arg
 740 745 750
 Thr Pro Ser Gln Glu Gln Leu Leu Ala Glu Ala Leu Ala Ala Glu
 755 760 765
 Gly Glu Asp Gly Val Ser Glu Ala Gln Glu Thr Pro Asp His Ala Ile
 770 775 780
 Phe Arg Trp Leu Ala Ile Ser Phe Ala Ser Ala His Leu Thr Met Thr
 785 790 795 800
 Glu Pro Tyr Arg Gly Gly Cys Gln Ala Gln Asp Tyr Thr Ser Gly Met
 805 810 815
 Gly Ile Val Asn Gly Ala Lys Trp Asn Pro Arg Ser Gly Thr Phe Asn
 820 825 830
 Asp Phe Ser Tyr Leu His Thr Asn Cys Leu Glu Leu Ser Val Tyr Ile
 835 840 845
 Asp Lys Phe Pro His Glu Ser Glu Leu Pro Arg Glu Trp Glu Asn Asn
 850 855 860
 Lys Glu Ala Leu Leu Thr Phe Met Glu Gln Val His Arg Gly Ile Lys
 865 870 875 880
 Gly Val Val Thr Asp Glu Gln Gly Ile Pro Ile Ala Asn Ala Thr Ile
 885 890 895
 Ser Val Ser Gly Ile Asn His Gly Val Lys Thr Ala Ser Gly Gly Asp
 900 905 910
 Tyr Trp Arg Ile Leu Asn Pro Gly Glu Tyr Arg Val Thr Ala Glu Gly
 915 920 925

Tyr Thr Ser Ser Ala Lys Ile Cys Asn Val Asp Tyr Asp Ile Gly Ala
 930 935 940
 Thr Gln Cys Asn Phe Ile Leu Ala Arg Ser Asn Trp Lys Arg Ile Arg
 945 950 955 960
 Glu Ile Leu Ala Met Asn Gly Asn Arg Pro Ile Leu Gly Val Asp Pro
 965 970 975
 Ser Arg Pro Met Thr Pro Gln Gln Arg Arg Met Gln Gln Arg Arg Leu
 980 985 990
 Gln Tyr Arg Leu Arg Met Arg Glu Gln Met Arg Leu Asn Ser Thr Ala
 995 1000 1005
 Gly Pro Ala Thr Ser Pro Thr Pro Ala Leu Met Pro Pro Pro Ser Pro
 1010 1015 1020
 Thr Pro Ala Ile Thr Leu Arg Pro Trp Glu Val Leu Pro Thr Thr Thr
 1025 1030 1035 1040
 Ala Gly Trp Glu Glu Ser Glu Thr Glu Thr Tyr Thr Glu Val Val Thr
 1045 1050 1055
 Glu Phe Glu Thr Val Phe Thr Asp Leu Glu Val Glu Glu Leu Glu Glu
 1060 1065 1070
 Glu Glu Glu Arg Glu Glu Glu Glu Met Asp Thr Gly Leu Thr Phe Pro
 1075 1080 1085
 Leu Thr Thr Val Glu Thr Tyr Thr Val Asn Phe Gly Asp Phe
 1090 1095 1100

<210> 5
 <211> 10
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: CArG box

<400> 5
 ccwwwwwggg

10

<210> 6
 <211> 30
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: CArG box (3X)

<400> 6
 ccwwwwwggg ccwwwwwggg ccwwwwwggg

30

<210> 7
 <211> 10

<212> DNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: modified oligo

<400> 7
 ccaaatatgg 10

<210> 8
 <211> 10
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: modified oligo

<400> 8
 ccacacatgg 10

<210> 9
 <211> 29
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: primer

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<210> 10
 <211> 26
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: primer

<400> 10
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<210> 11
 <211> 21
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: primer

<400> 11
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<210> 12
 <211> 21
 <212> DNA
 <213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: primer

<400> 12

ttaccacgat gttcctcttg a

21

<210> 13

<211> 21

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: primer

<400> 13

aggaaacacc aaggtcaagc a

21

<210> 14

<211> 20

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: primer

<400> 14

gggactgtac cacaggttag

20

<210> 15

<211> 26

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: primer

<400> 15

tgaaggctcg tgtgaacgga tttggc

26

<210> 16

<211> 24

<212> DNA

<213> Artificial Sequence

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catgtaggcc atgaggtcca ccac

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<210> 17

<211> 21

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: primer

<400> 17
 cagtggctgg aaagcaagag c 21

 <210> 18
 <211> 18
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> Description of Artificial Sequence: primer

 <400> 18
 gggctggggc agacgggc 18

 <210> 19
 <211> 45
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> Description of Artificial Sequence: CArG element

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 <210> 20
 <211> 17
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> Description of Artificial Sequence: primer

 <400> 20
 atctggttgt cctcaat 17

 <210> 21
 <211> 17
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> Description of Artificial Sequence: primer

 <400> 21
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 <210> 22
 <211> 26
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> Description of Artificial Sequence: EcoRI site

 <400> 22
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<210> 23
<211> 28
<212> DNA
<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: HindIII site

<400> 23

cgaagcttga agtccccaaa gttcactg

28

61
